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L15 same 12



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<i>DB=PGPB,USPT,USOC,EPAB,JPAB,DWPI; PLUR=YES; OP=ADJ</i>			
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<u>L5</u>	L4 with l3	1	<u>L5</u>
<u>L4</u>	recharging	31485	<u>L4</u>
<u>L3</u>	l2 with l1	153	<u>L3</u>
<u>L2</u>	cationic lipid or cationic amphiphile	7464	<u>L2</u>
<u>L1</u>	polyanio\$	9582	<u>L1</u>

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L16: Entry 6 of 68

File: PGPB

Nov 13, 2003

DOCUMENT-IDENTIFIER: US 20030212031 A1

TITLE: Stable lipid-comprising drug delivery complexes and methods for their production

Summary of Invention Paragraph:

[0008] This invention provides methods for producing lipid-comprising drug delivery complexes having a net positive charge and/or a positively charged surface. By "drug" as used throughout the specification and claims is meant any molecular entity, which is either monomeric or oligomeric, and which, when complexed with lipid or with lipid and polycation, is being administered to an individual for the purpose of providing a therapeutic effect to the recipient. Thus, macromolecules having an overall net negative charge or regions of negativity would be expected to be capable of forming the delivery complexes of this invention. Macromolecules which are particularly suitable for use with the complexes of this invention are for example, DNA, RNA, oligonucleotides or negatively charged proteins. However, macromolecules having a positive charge (e.g., large cationic protein) would also be expected to be capable of forming the complexes of this invention by sequentially complexing the cationic macromolecule with anionic molecule or polymer and then with cationic lipid.

Brief Description of Drawings Paragraph:

[0056] Thus, in addition to cationic lipids, cationic liposomes used to form the complexes of the invention may contain other lipids in addition to the cationic lipids. These lipids include, but are not limited to, lyso lipids of which lysophosphatidylcholine (1-oleoyl lysophosphatidylcholine) is an example, cholesterol, or neutral phospholipids including dioleoyl phosphatidyl ethanolamine (DOPE) or dioleoyl phosphatidylcholine (DOPC) as well as various lipophylic surfactants, containing polyethylene glycol moieties, of which Tween-80 is one example. The lipid complexes of the invention may also contain negatively charged lipids as well as cationic lipids so long as the net charge of the complexes formed is positive and/or the surface of the complex is positively charged. Negatively charged lipids of the invention are those comprising at least one lipid species having a net negative charge at or near physiological pH or combinations of these. Suitable negatively charged lipid species include, but are not limited to, CHEMS (cholesteryl hemisuccinate), NGPE (N-glutaryl phosphatidylethanolanine), phosphatidyl glycerol and phosphatidic acid or a similar phospholipid analog.

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L16: Entry 37 of 68

File: PGPB

Dec 13, 2001

DOCUMENT-IDENTIFIER: US 20010051610 A1

TITLE: Method for nucleic acid transfection of cells

Detail Description Paragraph:

[0158] An experiment was performed to demonstrate that ZnCl₂.sub.2 can be used to enhance the in vitro transfection activity of cationic lipid/nucleic acid complexes. To perform this study, cationic lipid/nucleic acid/zinc mixtures were prepared by mixing appropriate amounts of serum-free DMEM, cationic liposomes, zinc chloride and pCMV.FOX.Luc.2 plasmid DNA in a polystyrene tube. In this experiment, the cationic lipid/nucleic acid complexes were formed at different cationic lipid:nucleic acid phosphate charge ratios. Specifically, complexes were formed at charge ratios of 0.5, 0.75, 1.0, and 2.0. Complexes formed at charge ratios above 1.0 possess a net positive charge whereas those with a charge ratio below 1.0 have a net negative charge. The cationic lipid/nucleic acid phosphate charge ratio is an important experimental parameter that influences the transfection activity of cationic lipid/nucleic acid complexes. In many instances, complexes possessing a net positive charge are more active than those with a net neutral or net negative charge. Cationic lipid/nucleic acid complexes at each charge ratio were screened for transfection activity in NIH 3T3 cells in the presence of different concentrations of zinc chloride (0.0, 0.1, 1, 10, 100, and 1000 μM). NIH 3T3 cells are a murine fibroblast tissue culture cell line commonly used to demonstrate the in vitro transfection activity of gene delivery reagents. After 48 hours post-application of the cationic lipid/DNA/zinc solutions to the cells, the cells were lysed with lysis buffer and the lysate was assayed for luciferase specific activity. As illustrated in FIG. 3 and Table 10, the data clearly illustrates that zinc, when added to a cationic liposome/DNA mixture, can enhance in vitro transfection of cultured NIH 3T3 murine fibroblast cells by two to forty fold depending on the cationic lipid to nucleic acid charge ratio. The effect was more pronounced at lower charge ratios, however, and effect was observed at all the charge ratios screened. The ability of the methods of the present invention to increase the activity of low charge ratio cationic lipid/DNA complexes is highly advantageous over prior art systems because highly charged complexes have a significant amount of associated cytotoxicity.

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File: DWPI

Nov 14, 1991

DERWENT-ACC-NO: 1991-353894

DERWENT-WEEK: 199148

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TITLE: Intracellular delivery of biologically active cpds. - comprises self assembling lipid complexes

Basic Abstract Text (1):

(A) A method for making a vehicle for administering a bioactive substance to a cell is new. The method comprise (a) providing the substance in a first lipid vesicle which comprises at least one negatively charged lipid and having a net negative charge; and (b) combining first lipid vesicle with second lipid vesicles comprising at least one cationic lipid and having a net positive charge, so the first lipid vesicles are coated with at least one positively charged lipid vesicle.

Basic Abstract Text (2):

(B) A method of making a vehicle for administering a bioactive substance to a cell is also claimed and comprises: (a) contacting a bioactive substance (BS) with at least one lipid vesicle (LV) comprising a negatively charged lipid species and having a net negative charge, so the BS and LV form a first lipid complex; and (b) contacting first lipid complex with lipid vesicles comprising at least one cationic lipid and having a net positive charge where the first lipid complex and the positively charged lipid vesicles form a second lipid complex.

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END OF SEARCH HISTORY